

Product Sheet

H_GHR Reporter Cell Line

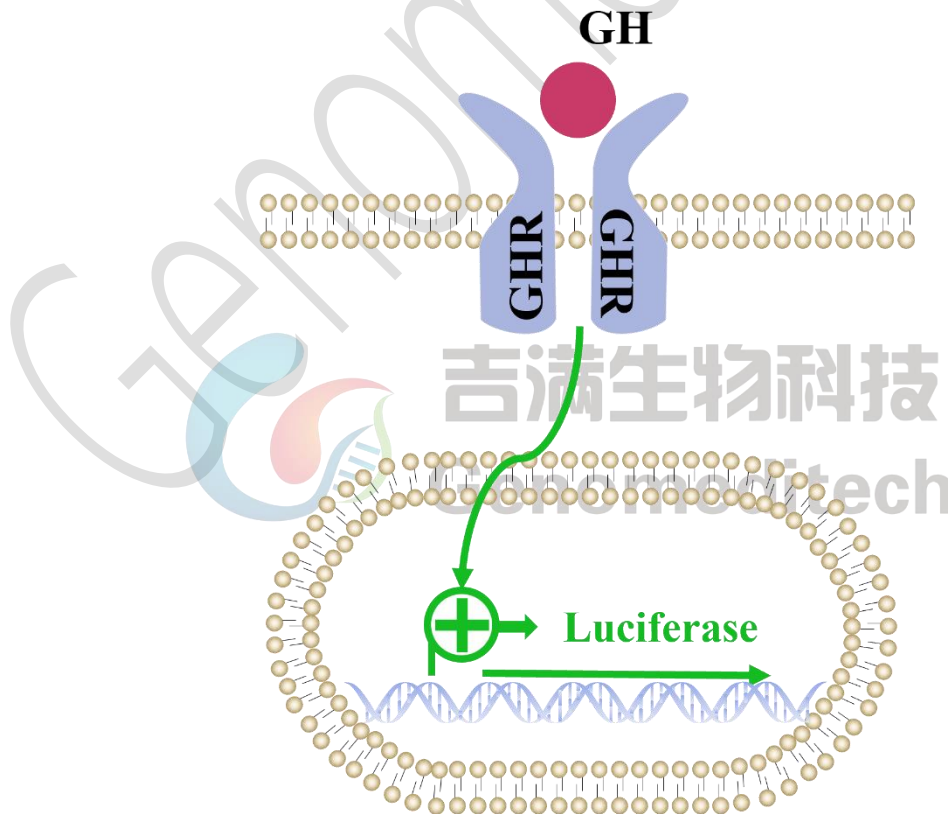
Catalog number: GM-C30425

Version 3.3.1.250103

Growth hormone (GH) is a polypeptide hormone secreted by the anterior pituitary gland that promotes growth, cell regeneration, and metabolic regulation. It is crucial during childhood and adolescence for stimulating bone and soft tissue growth, and it helps maintain body composition and metabolic balance in adults. GH secretion is regulated by factors such as growth hormone-releasing hormone (GHRH), growth hormone-inhibiting hormone (GHIH), and blood glucose levels.

GH signaling occurs through its binding to the growth hormone receptor (GHR), activating JAK2 (Janus kinase 2) and initiating the STAT5 (signal transducer and activator of transcription 5) pathway. This regulates gene expression, affecting cell growth, differentiation, and metabolism. GH also promotes the synthesis of insulin-like growth factor 1 (IGF-1), which further regulates cell proliferation and growth.

H_GHR Reporter Cell Line is a clonal stable cell line constructed using lentiviral technology, constitutive expression of the GHR gene, along with signal-dependent expression of a luciferase reporter gene. When GH binds to GHR, it activates downstream signaling pathways, leading to the expression of luciferase. The luciferase activity measurement indicates the activation level of the signaling pathway and can thus be used to evaluate the in vitro effects of drugs related to GHR.



Specifications

Quantity	5E6 Cells per vial, 1 mL
Product Format	1 vial of frozen cells
Shipping	Shipped on dry ice
Storage Conditions	Liquid nitrogen immediately upon receipt
Recovery Medium	RPMI 1640+10% FBS+1% P.S+20 ng/mL H_GH
Growth medium	RPMI 1640+10% FBS+1% P.S+20 ng/mL H_GH+5 µg/mL Blasticidin+0.25 µg/mL Puromycin
Note	None
Freezing Medium	90% FBS+10%DMSO
Growth properties	Suspension
Growth Conditions	37°C, 5% CO ₂
Mycoplasma Testing	The cell line has been screened to confirm the absence of Mycoplasma species.
Safety considerations	Biosafety Level 2
Note	It is recommended to expand the cell culture and store a minimum of 10 vials at an early passage for potential future use.

Materials

Reagent	Manufacturer/Catalogue No.
RPMI 1640	VivaCell/C3010-0500
Fetal Bovine Serum	Cegrogen biotech/A0500-3010
Pen/Strep	Thermo/15140-122
Recombinant Human GH	Novoprotein/C725
Blasticidin	Genomeditech/GM-040404
Puromycin	Genomeditech/GM-040401
GMOne-Step Luciferase Reporter Gene Assay Kit	Genomeditech/GM-040503

Figures

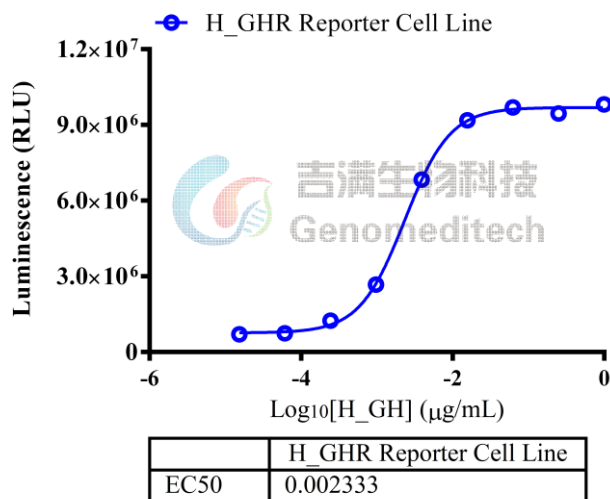


Figure 1 | Response to Recombinant Human GH. The H_GHR Reporter Cell Line (Cat. GM-C30425) at a concentration of 1E5 cells/well (96-well format) was stimulated with serial dilutions of Recombinant Human GH (Novoprotein/C725) in assay buffer (RPMI 1640 + 1% FBS + 1% P.S) for 24 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [14.0]. Data are shown by drug mass concentration.

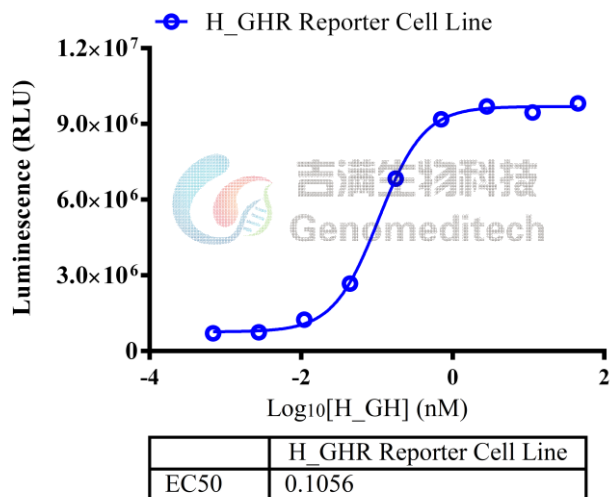


Figure 2 | Response to Recombinant Human GH. The H_GHR Reporter Cell Line (Cat. GM-C30425) at a concentration of 1E5 cells/well (96-well format) was stimulated with serial dilutions of Recombinant Human GH (Novoprotein/C725) in assay buffer (RPMI 1640 + 1% FBS + 1% P.S) for 24 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [14.0]. Data are shown by drug molar concentration.

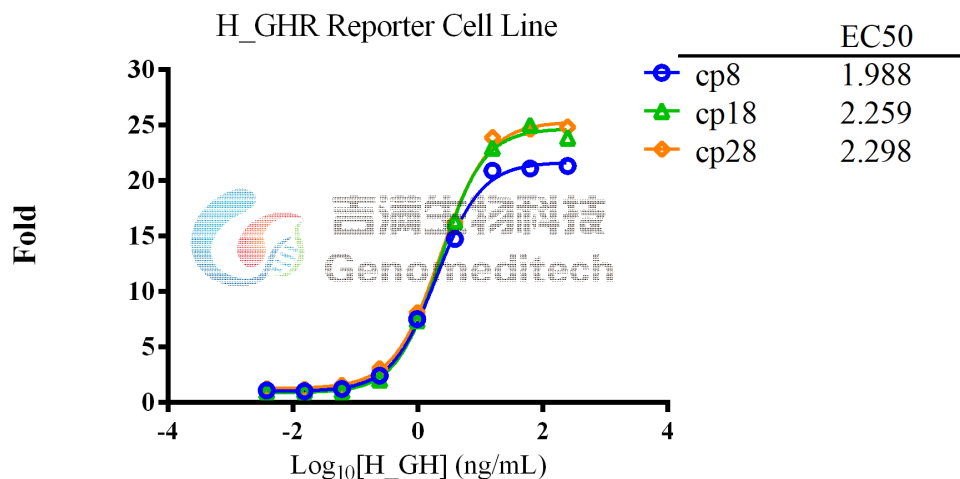


Figure 3 | The passage stability of response to Recombinant Human GH. The passage 8, 18 and 28 of H_GHR Reporter Cell Line (Cat. GM-C30425) at a concentration of 1E5 cells/well (96-well format) were stimulated with serial dilutions of Recombinant Human GH (Novoprotein/C725) in assay buffer (RPMI 1640 + 1% FBS + 1% P.S) for 24 hours. The firefly luciferase activity was measured using the GOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). Data are shown by drug mass concentration.

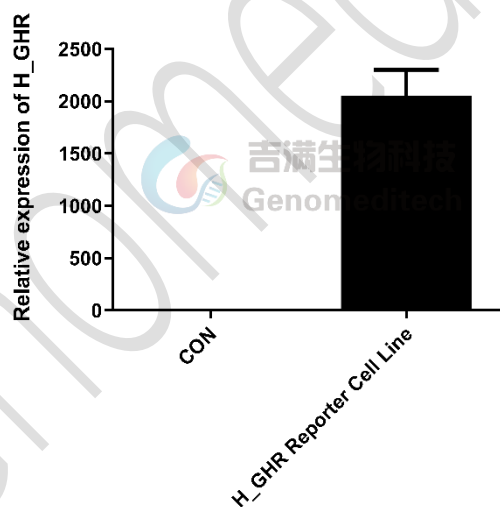


Figure 4 | The mRNA expression levels of H_GHR in the H_GHR Reporter Cell Line (Cat. GM-C30425) were determined by RT-Qpcr.

Cell Recovery

Recovery Medium: RPMI 1640+10% FBS+1% P.S+20 ng/mL H_GH

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70°C . Storage at -70°C will result in loss of viability.

- a) Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 - 3 minutes).
- b) Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions.
- c) Transfer the vial contents to a centrifuge tube containing 5.0 mL complete culture medium. And spin at approximately $176 \times g$ for 5 minutes. Discard supernatant.
- d) Resuspend cell pellet with the recommended complete medium. And dispense the suspension into 1-2 T-25 culture flasks.
- e) Incubate the culture at 37°C in a suitable incubator. A 5% CO_2 in air atmosphere is recommended if using the medium described on this product sheet.

Cell Freezing

Freezing Medium: 90% FBS+10%DMSO

- a) Centrifuge at $176 \times g$ for 3 minutes to collect cells.
- b) Resuspend the cells in pre-cooled freezing medium and adjust the cell density to 5×10^6 cells/mL.
- c) Aliquot 1 mL into each vial.
- d) Place the vials in a controlled-rate freezing container and store at -80°C for at least 1 day, then transfer to liquid nitrogen as soon as possible.

Cell passage

Growth medium: RPMI 1640+10% FBS+1% P.S+20 ng/mL H₂GH+5 $\mu\text{g/mL}$ Blasticidin+0.25 $\mu\text{g/mL}$ Puromycin

Approximately 48-72 hours after the initial thawing, the cells can be passaged for the first time. After this initial passage, the culture medium can be adjusted to growth medium supplemented with antibiotics. If cells are not passaged within 48 hours, it is recommended to add some fresh recovery medium and place the flask horizontally.

- a) When the cell density reaches $1 - 1.2 \times 10^6$ cells/mL, subculture the cells. Do not allow the cell density to exceed 1.4×10^6 cells/mL.
- b) It is recommended to use T-25 flasks for subculturing.
- c) These cells are suspension cells, and it is recommended to use the "half-medium change" method to maintain optimal cell conditions during passaging.
- d) During passaging, you can directly add fresh growth medium to the culture flask, gently pipette to resuspend the cells, and then transfer the cell suspension to a new T-25 flask for continued culture.

Subcultivation Ratio: Maintain cultures at a cell concentration between 3×10^5 and 1×10^6 viable cells/mL.

Medium Renewal: Every 2 to 3 days

Notes

- a) These cells are sensitive to density, so please ensure that the cell density is maintained within an appropriate range during culture and subculturing.
- b) During the first passage, pay attention to the nutrient supply; if not subculturing, make sure to add fresh recovery medium every other day as needed.

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